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BIAS ASSOCIATED WITH FOOD ANALYSIS IN GIZZARDS OF BLUE-WINGED TEAL

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Abstract: The food contents of 67 adult and flying juvenile blue-winged teal (*Anas discors*) collected during the spring and summer months of 1967 and 1968 on prairie wetlands in central North Dakota were statistically compared. The results demonstrated disagreement in food composition between the esophageal and gizzard contents. Differences observed in wild birds were confirmed experimentally in 13 feeding trials in which drake blue-winged teal were fed natural foods and then killed at time intervals ranging from 2.5 minutes to 72 hours later. The magnitude of the bias related to digestibility increased in direct proportion to the time lapse between feeding and sampling. Soft foods broke down within minutes while hard seeds were retained for days. Reliable information can be obtained by analyses of esophageal contents, collecting birds that are actively feeding, immediately removing contents of the esophagus, and preserving foods in 80 percent alcohol.

A reliable assessment of the kind of foods consumed is essential in understanding species ecology and in assessing the ecological threats posed by human activities. Stickle (1969:24), commenting on environmental assessment and research needs, stated, "Investigations should be perceptive, rather than general; they should focus on the specific habitats and on the food organisms actually consumed in these habitats." If this end is to be accomplished, bias must be avoided and the organism's entire life history considered.

A preliminary investigation in 1967 of foods ingested by spring- and summer-collected blue-winged teal on the prairie wetlands of central North Dakota demonstrated marked differences in species composition between food items contained in the esophagus and gizzard. Perret (1962) demonstrated a similar disagreement in mallards (*Anas platyrhynchos*); Bartonek and Hickey (1969) demonstrated disagreement among the contents of the esophagus, proventriculus, and gizzard of three species of diving ducks; Dillon (1957) described a similar situation in foods consumed by waterfowl on wintering areas; and Dirsehl (1969:79) reported that gizzard contents inflate the importance of hard-seeded plants

in the diet of lesser scaup (*Aythya affinis*) and blue-winged teal. Moyle (1961:2), commenting on differential digestive rates of waterfowl foods, stated, "It seems highly probable that soft-bodied invertebrates like chironomids or snails would be identifiable in the gizzard for a much shorter period after ingestion than hard-shelled pondweed fruits. Differential digestive rates of food materials by ducks certainly merits investigation."

Although concern has been expressed over the bias associated with determination of the kinds of foods consumed by waterfowl as determined by gizzard analysis, attempts (Bartonek 1968) have been made to investigate different rates of digestion on an experimental basis. This paper contains the combined results of two complementary studies of blue-winged teal: (1) an examination of the food eaten by birds collected in the wild and (2) feeding trials with wild, captive-reared birds. The purpose of this paper is (1) to ascertain the bias associated with food found in the gizzard; (2) to explain why bias occurs; and (3) to suggest procedures that can be used to avoid bias. The data presented are part of a larger study dealing with the feeding ecology of waterfowl and designed to pro-

vide information that can be used to assess the significance of environmental changes occurring in waterfowl breeding habitat.

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METHODS

In the first phase of the study the contents of the esophagi and gizzards of 67 adult and flying juvenile blue-winged teal, collected during the spring and summer of 1967 and 1968, were compared statistically by means of the paired *t* test (Steel and Torrie 1960:79). The percentage of animal material contained in the esophagus was compared to that found in the gizzard. Percentages were calculated from the total volume of foods contained in each of the two anatomic parts. The contents of the proventriculus were also measured but produced results that were intermediate between those of the esophagus and gizzard; therefore the contents of this zone are not considered in this paper. Prior to statistical analysis, the data were subjected to square root transformation, $\sqrt{x + 1}$ (Steel and Torrie 1960).

Birds were collected while actively feeding on 39 different wetlands located in central North Dakota ranging from fresh-temporary, to permanent-saline water areas. Food items were removed immediately and preserved in 80 percent ethyl alcohol to minimize postmortem digestion and autolysis as discussed by Koersveld (1950), Dillery (1965), and Bartonek (1968). Food items were counted in the laboratory, external moisture removed, and volumes measured by liquid displacement. Only the

birds containing a total volume of food in the esophagus exceeding 0.05 ml were included in this comparison.

The second phase of the study included 13 feeding trials, each consisting of identical feedings of pairs of 7-week-old drake blue-winged teal which were subsequently sampled over a series of time intervals ranging from 2.5 minutes to 72 hours. The animal and plant items fed in each trial were selected from a series of natural foods that comprised the diet of wild teal and ranged in digestibility from the soft amphipod (*Hyalella azteca*) to the hard achenes of bulrushes (*Scirpus* spp.). Food items included four species of plant seeds: hardstem bulrush (*S. acutus*), alkali bulrush (*S. pululosus*), slough sedge (*Carex athetoides*) and western waterplantain (*Alisma triculale*); and three invertebrate species representing Insecta (*Chironomus* spp.), Crustacea (*H. azteca*), and Mollusca (*Gymnulus* spp.). *S. pululosus* was fed in the first nine trials and *S. acutus* in the remaining four.

Food items (25 of each) were inserted approximately 50 mm into the duck's esophagus by using a modified 1 cc disposable plastic syringe (7 mm o.d., 5 mm i.d.). Two syringes were used; each contained 75 food items. Dry seeds were deposited first followed by live invertebrates. At the end of each predetermined time interval, birds were killed and immediately dissected. Ligatures were secured at the anterior end of the esophagus and at the junctions of the esophagus and proventriculus, proventriculus and gizzard, gizzard and intestine, and intestine and cloaca. The alimentary tract and fecal materials were washed, screened, and examined for force-fed food items. All items were counted if sufficiently intact to insure accurate enumeration. Birds were placed, during

Table 1. A comparison of the percent by volume of animal material found in the esophagus and gizzard of blue-winged teal collected while feeding in May-June and August-September, 1967 and 1968, in North Dakota (flying juveniles were included in the latter sample).

| PARAMETER | MAY-JUNE | | AUGUST-SEPTEMBER | |
|--|-----------|---------|------------------|---------|
| | Esophagus | Gizzard | Esophagus | Gizzard |
| Sample size | 30 | 30 | 31 | 31 |
| Mean food volume (ml) | 0.97 | 0.30 | 1.21 | 0.19 |
| Mean percent (animal material) | 91.16 | 32.09 | 80.58 | 29.23 |
| Mean confidence limits (animal material—0.05 level) | ± 4.50 | ± 8.00 | ± 11.55 | ± 10.24 |
| Calculated <i>t</i> value for difference of means (animal material) | 12.108*** | | 8.468*** | |

*** $P < 0.001$.

each feeding trial, in individual dry 10-gal polyethylene aquaria to insure recovery of fecal materials. Water was supplied throughout the study. Supplemental food in the form of a custom-prepared waterfowl crumble diet (19 percent protein) was supplied ad libitum for all trials that exceeded 2 hours, with the exception of one 12-hour trial during which food was inadvertently withheld.

Birds used in this second phase were from wild stock artificially incubated and hatched during the first 2 weeks in July. The diet prior to experimental feeding consisted of crushed starter pellets (29 percent protein) for 3 weeks, grower pellets (19 percent protein) for 2 weeks, and a mixture of the grower pellets, whole wheat, and whole barley for 2 weeks. Grit was provided as a mixture of fine gravel and sand. The study was initiated on August 30 and terminated on September 21, with the exception of the 72-hour trial which was delayed until November, 1968.

Invertebrates were never found in the gizzard after the 20-minute trial and were excluded in trials that exceeded 2 hours. The information obtained from paired birds within individual time trials was combined to simplify presentation.

RESULTS

A statistical comparison of the esophageal and gizzard contents of teal collected in spring and summer is presented in Table 1. The calculated *t* values for the spring and summer birds were 12.11 and 8.47, respectively. Therefore the null hypothesis was rejected, namely foods identified from the esophagus and gizzard were not identical.

The detailed results of the first three feeding trials (2.5–10 minutes) are presented in Table 2. The transfer of food items from the esophagus through the proventriculus to the gizzard occurred quite rapidly. The amphipods and snails broke down rapidly in the gizzard. At the end of 10 minutes, 100 percent of the amphipods, 82 percent of the snails, and 24 percent of the diptera larvae were digested beyond distinction.

A summary of the results of the 13 trials is presented in Table 3. The number of western waterplantain nutlets remained relatively stable for 20 minutes, then steadily declined to the 2-hour series after which these nutlets no longer appeared in the gizzard. The number of sedge achenes remained relatively stable for 2 hours, then declined to the 24-hour trial, and dis-

Table 2. Movement of force fed food items through the digestive tract. Data are the number of individual items retained by paired blue-winged teal at the termination of each time trial.

| FOOD ITEMS | NUMBER FED (two birds) | Volume Fed (ml) | NUMBER OF FOOD ITEMS FED AND RELOCATED ^a | | | TOTAL IDENTIFIED | UNACCOUNTED FOR |
|--------------------------|---------------------------|--------------------|---|----------------|---------|---------------------|--------------------|
| | | | Esophagus | Proventriculus | Gizzard | | |
| 2.5-Minute Trial | | | | | | | |
| <i>Hyalella azteca</i> | 50 | 0.00 | 50 | 6 | 14 | 50 | 0 |
| <i>Cyranulus</i> spp. | 50 | 0.35 | 35 | 8 | 7 | 50 | 0 |
| <i>Chironomus</i> spp. | 50 | 1.30 | 50 | 0 | 0 | 50 | 0 |
| <i>Alisma triviale</i> | 50 | 0.01 | 36 | 7 | 7 | 50 | 0 |
| <i>Carex atherodes</i> | 50 | 0.00 | 31 | 6 | 10 | 50 | 0 |
| <i>Scirpus paludosus</i> | 50 | 0.10 | 32 | 3 | 15 | 50 | 0 |
| Total (2 birds) | 300 | 2.54 | 217 | 30 | 53 | 300 | 0 |
| Percent of Total | 100 | 100 | 72 | 10 | 18 | 100 | 0 |
| 5-Minute Trial | | | | | | | |
| <i>Hyalella azteca</i> | 50 | 0.00 | 2 | 0 | 10 | 12 | 38 |
| <i>Cyranulus</i> spp. | 50 | 0.35 | 4 | 1 | 20 | 25 | 25 |
| <i>Chironomus</i> spp. | 50 | 1.30 | 7 | 0 | 43 | 50 | 0 |
| <i>Alisma triviale</i> | 50 | 0.01 | 13 | 2 | 31 | 46 | 4 |
| <i>Carex atherodes</i> | 50 | 0.00 | 4 | 0 | 45 | 49 | 1 |
| <i>Scirpus paludosus</i> | 50 | 0.10 | 2 | 1 | 47 | 50 | 0 |
| Total (2 birds) | 300 | 2.54 | 32 | 4 | 196 | 232 | 68 |
| Percent of total | 100 | 100 | 11 | 1 | 65 | 77 | 23 |
| 10-Minute Trial | | | | | | | |
| <i>Hyalella azteca</i> | 50 | 0.00 | 0 | 0 | 0 | 0 | 50 |
| <i>Cyranulus</i> spp. | 50 | 0.35 | 0 | 1 | 8 | 9 | 41 |
| <i>Chironomus</i> spp. | 50 | 1.30 | 1 | 1 | 30 | 38 | 12 |
| <i>Alisma triviale</i> | 50 | 0.01 | 1 | 1 | 43 | 45 | 5 |
| <i>Carex atherodes</i> | 50 | 0.00 | 1 | 0 | 48 | 49 | 1 |
| <i>Scirpus paludosus</i> | 50 | 0.10 | 1 | 0 | 49 | 50 | 0 |
| Total (2 birds) | 300 | 2.54 | 4 | 3 | 164 | 191 | 109 |
| Percent of total | 100 | 100 | 1 | 1 | 62 | 61 | 30 |

^a No food items found in intestine and feces.

appeared in subsequent trials. Bulrush achenes remained in the gizzard throughout the entire time series, with a moderate reduction (26 percent) in numbers occurring after 2 hours. As the number of bulrush achenes disappeared from the gizzard, the number in the lower digestive tract and feces increased (Table 3). Twenty-four percent of the achenes fed were found whole in the lower intestine and fecal material at the termination of the

24-hour trial. Fifty-eight percent of the achenes still remained in the gizzard; this left only 18 percent unaccounted for.

The variation in the number of food items retained between birds within individual time trials remained relatively low. The mean difference for the 13 time trials, consisting of 150 items fed to each bird, was 8.9 food items, and the maximum difference was 19 items. It is reasonable to assume that under natural conditions the

Table 3. Differential retention rates of force-fed food items. Data expressed as the sum of individual items retained by paired blue-winged teal at the termination of each of 13 time trials.

| NUMBER OF FOOD ITEMS RETAINED IN PAIRED BIRDS AT THE TERMINATION OF EACH TIME TRIAL. | | | | | | | | | | | | | | |
|--|--------|---------|-----|-----|-----|-----|----|----|-------|----|----------------|----|----|----|
| Food Items | Time / | Minutes | | | | | | | Hours | | | | | |
| | | 2.5 | 5 | 10 | 20 | 30 | 40 | 50 | 1 | 2 | 12 | 24 | 48 | 72 |
| Upper digestive tract (esophagus, proventriculus and gizzard) | | | | | | | | | | | | | | |
| <i>Hyalella azteca</i> | 50 | 12 | 0 | 3 | 0 | 0 | 0 | 0 | 0 | 0 | — ^a | — | — | — |
| <i>Cyranulus</i> spp. | 50 | 25 | 0 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | — | — | — | — |
| <i>Chironomus</i> spp. | 50 | 50 | 38 | 17 | 0 | 0 | 0 | 0 | 0 | 0 | — | — | — | — |
| <i>Alisma tritiale</i> | 50 | 40 | 45 | 41 | 20 | 33 | 3 | 7 | 5 | 0 | 0 | 0 | 0 | 0 |
| <i>Carex altherodes</i> | 50 | 40 | 40 | 47 | 43 | 48 | 28 | 40 | 47 | 5 | 1 | 0 | 0 | 0 |
| <i>Scirpus</i> spp. | 50 | 50 | 40 | 40 | 50 | 47 | 42 | 44 | 37 | 20 | 20 | 10 | 14 | 14 |
| Total (2 birds each time trial) | 300 | 232 | 191 | 158 | 113 | 128 | 73 | 97 | 89 | 31 | 30 | 10 | 14 | 14 |
| Lower digestive tract (intestine and feces) | | | | | | | | | | | | | | |
| <i>Hyalella azteca</i> ^b | | | | | | | | | | | — | — | — | — |
| <i>Cyranulus</i> spp. ^b | | | | | | | | | | | — | — | — | — |
| <i>Chironomus</i> spp. | 0 | 0 | 0 | 0 | 0 | 0 | 1 | 0 | 0 | — | — | — | — | — |
| <i>Alisma tritiale</i> | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | — | — |
| <i>Carex altherodes</i> | 0 | 0 | 0 | 0 | 0 | 2 | 0 | 0 | 3 | 0 | 0 | 1 | 0 | 0 |
| <i>Scirpus</i> spp. | 0 | 0 | 0 | 0 | 0 | 0 | 1 | 0 | 4 | 3 | 12 | 7 | 7 | 7 |
| Total retained (2 birds in each time trial) | 0 | 0 | 0 | 1 | 0 | 2 | 2 | 0 | 7 | 3 | 12 | 8 | 7 | 7 |

^a Dash indicates item was excluded from feeding trial.^b Organisms never located in the lower digestive tract.

dominant foods consumed have a considerable influence on the breakdown and subsequent retention of hard seeds. In this case crushed food pellets approximated the soft animal foods that dominate the diet of blue-winged teal during the spring and summer months in central North Dakota (Table 1).

DISCUSSION

As the time lapse between feeding and sampling increased, the magnitude of the bias associated with the gizzard contents increased in direct proportion. Soft foods broke down within minutes while hard items were retained for several days.

Hard seeds in the gizzard must exert an abrasive action on soft foods and probably function as grit when soft foods dominate

the diet. Jordan and Hellrose (1951) reported that the effect of ingested lead was increased by harder foods in the diet. Beer and Tidyman (1942) presented evidence that gallinaceous birds substitute hard seeds for grit when grit levels fall below minimum values. Dement'ev and Gladkov (1967:499-500), describing foods ingested by garganey teal (*Anas querquedula*), reported that hard seeds frequently serve as gastroliths. The breakdown rate of hard seeds may vary, however, depending upon the length of time they are exposed to moisture prior to consumption; that is, the seeds become softer prior to germination.

The nutritional importance, in the diets of ducks, of plants having hard-coated seeds is minimized because some of these seeds are undigested and voided in the feces.

Voiding of these undigested and often viable seeds may serve as a primary means of dispersal for some species of aquatic and emergent plants (Bartonek 1968:107).

The significance of the disagreement among the contents of the esophagus, proventriculus, and gizzard is brought out by the work of Bartonek (1968), who determined that 95 percent of 125 studies of waterfowl food habits published from 1901 through 1965 were based wholly or in part upon food materials from the gizzard or stomach. Studies of this type tend to bias data in favor of items that are most difficult to digest, especially hard-coated seeds.

Adjusting data to compensate for different rates of digestion in the gizzard would be difficult under conditions where a wide spectrum of foods varying in resistance is consumed. Soft foods such as cladocera, even when consumed in large numbers, were never abundant in the gizzard. For example, the digestive tract of a hen gadwall (*A. strepera*) that had fed exclusively on cladocera carrying resistant eggs (ephippia) was examined, and the proportion of female cladocera carrying eggs was 1 in 5.2. This ratio remained relatively constant in the esophagus and proventriculus. Only 14 cladocera could be found in the gizzard, however, as compared to 225 ephippia, suggesting that proportionately 1,180 cladocera had previously entered the gizzard and had been digested. Microcrustaceans were readily identified when the contents of the esophagus were removed and preserved in alcohol.

Bartonek (1968:100) studied the differential breakdown of force-fed foods in 3½-week-old mallards. The overall breakdown rate of soft vs. hard foods followed a pattern similar to that described for blue-winged teal. In his study, cladocera broke down within 15 minutes; all animal ma-

terials broke down in less than 1 hour. Thirty-five percent of the hardstem bulrush achenes remained to the end of the study at 24 hours. Although the breakdown sequence reported was relatively the same, the 7-week-old teal in our study digested animal materials more rapidly than did the 3½-week-old mallards in Bartonek's study. This probably can be attributed in part to the age of the two species and the resulting differences in the size and development of their gizzards. Malone (1965) demonstrated that the rate of passage of brine shrimp (*Artemia salina*) eggs through the digestive tract of mallards was related to the hardness of other foods consumed. Invertebrate eggs fed with soft plants and animals passed rapidly while hard foods retarded passage, suggesting that hard foods are retained in the gizzard while soft foods are processed rapidly and passed into the intestine.

During the 1967 North Dakota teal season, 110 birds obtained from hunters' bags were examined. Of these only 33 (30 percent) contained food in the esophagus. Lack of food in the esophagus may have been caused by disturbance during hunting, but the rapid rate of movement of foods into the gizzard and the tendency for birds to rest following feeding are also contributing factors. Blue-winged teal flying into decoys or over a pass would seem less likely to contain foods in the esophagus than would birds obtained by jump-shooting.

Birds killed by shooting occasionally regurgitated gizzard contents as was evidenced by the polished condition of seeds, sand grains, and mollusc shells found in the proventriculus and esophagus. Regurgitation was observed most often when the esophagus was empty or nearly so. A full esophagus appeared to retard the regurgi-

tation of gizzard contents. We never observed regurgitation, as Malone (1960:227) reported for two mallards, in birds killed with sodium pentobarbital, or under natural feeding conditions.

The esophagi of some of the birds examined in the field were filled to capacity with food. This condition appears to be related to the abundance of available food which permits the rate of consumption to exceed the capacity of the gizzard to process food materials. This condition, however, did not occur in every case as some birds were observed to feed actively for a period of 20 minutes without accumulating a large amount of food in the esophagus. In these cases, low food abundance effectively reduced the rate of consumption to a point not greatly exceeding the receptive capacity of the proventriculus and gizzard.

CONCLUSIONS

Disagreement in composition between the esophageal and gizzard contents in blue-winged teal relates to different breakdown rates determined by the resistance of individual food items to digestive juices, to the grinding action of the gizzard, and to the other foods consumed. This bias can be avoided, when investigating food consumption, by limiting investigation to esophageal contents (Bartonek 1968, Perret 1962). This was accomplished effectively in blue-winged teal by restricting sampling to birds that were actively feeding, removing the esophageal contents immediately (Bartonek and Murdy 1970), and preserving them in 80 percent ethyl alcohol.

The accuracy of information derived from hunter-killed birds can be increased if cooperation can be enlisted to process esophagi in a precise manner before post-mortem digestion occurs. Not only must

the time between death, dissection, and preservation be considered, but also the possibility of regurgitation which apparently is caused by agonial spasms related to crippling. Since regurgitation is related to an empty esophagus and is easily recognized by the polished condition of some regurgitated items, it does not pose a serious problem.

A bird that has been observed to feed for a period of time does not always contain an esophagus filled to capacity, but a volume adequate for analysis is usually present. The amount in the esophagus appears to be influenced by the relative abundance of food in the environment and the rate that the gizzard is able to process the food material being consumed. Soft materials therefore can be processed and used at a more rapid rate than hard foods.

Deficiencies associated with data derived from slain birds have been pointed out by Kharin and Tashchilin (1953), Madsen (1954) and others. Our data indicate that reliable information can be obtained if (1) effort is exerted to obtain actively feeding birds, (2) examination is restricted to the esophagus to avoid foods subjected to physical or chemical breakdown, and (3) food items are removed and preserved immediately to avoid post-mortem digestion.

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